AMENDMENTS TO THE CLAIMS

This listing of claims will replace all prior versions and listings of claims in the application:

Claims 1-19. (Canceled)

Claim 20. (Currently amended) A process for the preparation of ~~thrombolytic enzyme, named~~ Thrombinase, having a molecular weight in the range of 31,000 to 32,000 Daltons, which comprises:

(i) Culturing a ~~the~~ filtrate of cells of Bacillus sphaericus serotype H5a 5b in a culture medium consisting of 0.03 to 1.5% of yeast extract, 0.2 to 1.5% peptone, 1 to 1.6% sodium acetate, 0.3 to 0.5% beef extract, 0.2 to 0.5% sodium chloride, 0.5 to 1 % Soya peptone, and 0.68% ammonium sulphate at a pH in a ~~the~~ range of 7.2 to 8,

(ii) Removing the cultured cells by cross flow filtration using 0.22 mμ filter to obtain a cell supernatant,

(iii) Subjecting the cell supernatant ~~thus obtained~~ to two step ultra filtration:

a. with a first ultra filtration of the cell supernatant using 100,000 MW (Molecular Weight) cut off membrane to obtain a filtrate, and

b. with a second ~~followed by~~ ultra filtration of the filtrate ~~thus obtained,~~ using 10,000 MW cut off membrane to obtain a retentate,

(iv) Salting out the retentate with ammonium sulphate in a concentration in the range of 20 to 40% to obtain a precipitate,

(v) Subjecting the ~~resulting~~ precipitate to dialysis,

(vi) Re-precipitating the dialyzed precipitate using ice-cold acetone,

(vii) Reconstituting the re-precipitated precipitate in buffer,

(viii) Decolorizing the reconstituted precipitate by using modified CDR (Cell Debris Remover) treatment by eluting with a buffer containing 0.1 to 0.5 M NaCl and then dialyzing and lyophilizing,

(ix) Purifying the lyophilized precipitate ~~firstly~~ by ion exchange chromatography and ~~followed~~ by gel filtration chromatography to obtain a fraction showing fibrinolytic activity, and

(x) Dialyzing the fraction showing fibrinolytic activity and lyophilizing to obtain purified Thrombinase having a molecular weight in the range of 31,000 to 32,000 Daltons.

Claim 21. (Currently amended) A process as claimed in claim 20 wherein the buffer used in step (vii) is Tris 0.01 M and the pH is 8.0.

Claim 22. (Currently amended) A process as claimed in claim 20 wherein the amount of ice-cold acetone and crude enzyme used in step (vi) are in the ratio of 1:1 to 1:1.5 (v/v).

Claim 23. (Currently amended) A process as claimed in claim 21 wherein the amount of ice-cold acetone and crude enzyme used in step (vi) are in the ratio of 1:1 to 1:1.5 (v/v).

Claim 24. (Canceled)

Claim 25. (New) A process for the preparation of Thrombinase, having a molecular weight in the range of 31,000 to 32,000 Daltons, which comprises:

(i) Culturing a filtrate of cells of Bacillus sphaericus serotype H5a 5b in a culture medium consisting of 0.03 to 1.5% of yeast extract, 0.2 to 1.5% peptone, 1 to 1.6% sodium acetate, 0.3 to 0.5% beef extract, 0.2 to 0.5% sodium chloride, 0.5 to 1 % Soya peptone, and 0.68% ammonium sulphate at a pH in a range of 7.2 to 8,

(ii) Removing the cultured cells by cross flow filtration using 0.22 mμ filter to obtain a cell supernatant,

(iii) Subjecting the cell supernatant to two step ultra filtration:

a. with a first ultra filtration of the cell supernatant using 100,000 MW (Molecular Weight) cut off membrane to obtain a filtrate, and

b. with a second ultra filtration of the filtrate using 10,000 MW cut off membrane to obtain a retentate,

(iv) Salting out the retentate with ammonium sulphate in a concentration in the

range of 20 to 40% to obtain a precipitate,

(v) Subjecting the precipitate to dialysis,

(vi) Re-precipitating the dialyzed precipitate using ice-cold acetone,

(vii) Reconstituting the re-precipitated precipitate in buffer, and

(viii) Decolorizing the reconstituted precipitate by using modified CDR (Cell Debris Remover) treatment by eluting with a buffer containing 0.1 to 0.5 M NaCl and then dialyzing and lyophilizing.